### DEVELOPMENT OF OIL BIOREMEDIATION RESEARCH ON MARINE ENVIRONMENT IN INDONESIA

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#### ABSTRACT

Indonesian marine environment is one of the most vulnerable waters from oil pollution in the world. Therefore, a hard effort to minimize oil pollution impact is really needed. Physical and chemical approaches have been already popular to combat oil pollution; one of the other promising techniques is bioremediation, the use of microorganism to detoxify or remove pollutants. Research and application of bioremediation on soil environment has been started in Indonesia; however, in marine environment it is still need to be studied due to more complicated aspects and also difficulties. Development of bioremediation study on marine environment in Indonesia has been started from enumeration, isolation and identification of oil degrading (hydrocarbonoclastic) bacteria. Researches on taxonomic and functional genes have been conducted. Biostimulation and bioaugmentation studies are ongoing research which conducted from laboratory scale and microcosm scale to field experiment (sand column). To have a manual or guidelines on conducting bioremediation in marine environment is not easy and still many steps have to be done. Several aspects concerning with this study such as the diversity of polluted sites characteristic, oils characteristic, oceanographic conditions and engineering has to be studied comprehensively.

Key words: bioremediation, Indonesia, oil, marine.

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### INTRODUCTION

Indonesian marine waters is one of the most vulnerable environments from oil pollution in the world, especially Malacca, Lombok, Sunda, Makassar Straits and Coast of South Java (Indian Ocean) because Indonesian marine waters is a main road of world oil transportation and locations of more than 82 onshore and offshore exploration activities. Concerning with these activities, there were about 100.000 vessels and 32.836 tankers (capacity of 500.000 – 5.000.000 ton) passing through Malacca Strait during 1993 and for this time being, of course, it will be higher. There were also over 2000 kilometres of submersible pipes lines and hoses, over 150 oil jetties, 6 major processing plants and 4

major big Transit Terminals all over Indonesia. With this condition, oil spill accidents were recorded for at least 30 times in all over Indonesian marine waters during 1975 – 2008. Therefore, hard effort to minimize oil pollution impact is really needed. Beside physical and chemical approaches that are already popular, one of the other promising techniques is bioremediation.

The process of bioremediation is defined as the use of microorganism to detoxify or remove pollutants, which relies upon microbial enzymatic activities to transform or degrade the offending contaminants. It is an evolving method for the destruction removal and of manv environmental pollutants (Philp et al, 2005).

Research application and of bioremediation on soil environment has been started in Indonesia (Aditiawati et al., 2001; Sugoro and Aditiawati, 2003); however, in marine environment it is still need to be studied due to more complicated aspects and also difficulties. Therefore, comprehensive study in biodegradation processes that involve various factors such as pollutants. microorganisms, energy sources, physical and chemical conditions, toxicity, competition, and metabolites accumulation have to be done.

# CURRENT RESEARCH AND DEVELOPMENT

Development of bioremediation study on marine environment in Indonesia was started from enumeration, isolation and identification of oil degrading (hvdrocarbonoclastic) bacteria by Research Center for Oceanography-LIPI 1978. in Thavib (1978)showed the distribution of hydrocarbonoclastic bacteria in Jakarta Bay,

whereas Darmayati (2003, 2004) exhibited in Makassar Strait and Java Sea. It showed that oil degrading bacteria were present in the marine environment. By using conventional method, some hydrocarbonoclastics bacteria of Jakarta Bay have been isolated such as Aeromonas sp., Pseudomonas sp., Bacillus sp., B. megaterium and Corynebacterium sp. After that, similar study has been done in Makassar Strait, Malacca Strait and some other Indonesia marine waters by several researchers from different Indonesian Institution (LIPI, University of Riau and University of Hasanudin). Feliatra (1999) reported that Acinetobacter, Arthrobacter, Micrococcus and Bacillus of Malacca Strait have been isolated. Darmayati (2003) showed the distribution of hydrocarbonoclastic bacteria in Makassar Strait and Java Sea. Susilawati (2000)mentioned that Pseudomonas cepacia and P. gladioli were present in Makasar Strait. whereas Achromobacter putrefaciens, Acinetobacter haemolyticus and Vibrio alginolyticus of Java Sea were isolated (unpublished data).

Different approach with previous study on isolation and identification of oil degrading bacteria in Indonesia by using biomolecular has been done since 2005, after Collaboration Research between LIPI and NITE (Indonesia-Japan) was implemented. Darmayati et al., isolated 131 different (2008)hvdrocarbonoclastic bacterial strains of Jakarta Bay and Seribu Island. Some isolates (11 strains) were indicated as new strains. Common genus found was Alcanivorax, Marinobacter, Bacillus and Achromobacter. Further study in Jakarta ports showed that hydrocarbonoclastic bacteria of those sites consisted of 37 strains fluoranthene degrading bacteria, 30 strains crude oil degrading bacteria. 25 strains fluorene degrading bacteria, 9 strains phenanthrene degrading

bacteria, 9 strains naphtalene degrading bacteria, 8 strains dibenzothiophene degrading bacteria and 7 strains pyrene degrading bacteria (Darmayati, 2008).

Taxonomic studies on oil degrading bacteria have been conducted by Research Center for Biology-LIPI since 2005 in with NITE-LIPI Cooperation accordance These included morphology, project. physiology, chemotaxonomy and phylogeny. Sudiana, et al., (2009) mentioned that hydrocarbonoclastic bacteria of Jakarta Bay and Seribu Island isolated consisted of 5 groups namely alpha-proteobacteria, gamma-proteobacteria, Flavobacteria and Actinobacteria. Suspected Novel genus belonging to Rhodobacteriales has been found in 11 strains. One of them was a new species within *Rhodovulum*. Based on the taxonomic data obtained in this study, a novel species, Salipiger pariensis sp.nov, is proposed; RI-P1 LIPIMC is the type strain for Salipiger pariensis.

Functional genes have also been studied in this collaboration research by Research Center for Biotechnology-LIPI. These included screening functional genes for oil degradation and surfactant properties analyses of selected bacteria. The results showed that some potential isolates were able to degrade both alkene and poly-aromatic hydrocarbon. Sequences of mono-oxygenase gene from eight isolates showed 60-90% identity with alkane mono-oxygenase from public database of gene bank. This result showed the diversity of alkenes monooxygenase sequence from bacteria with different taxon (Yopi, 2009). Biodiversities of surfactant bacteria of Jakarta Bay and Pari Island were very divers and interesting (Susilaningsih, 2009).

Various approaches to do bioremediation are described as in situ and ex

situ methods. Those which may be involved are biostimulation, bioaugmentation, and natural monitored bioattenuation (Philp and 2005). Biostimulation Atlas. and bioaugmentation studies were studied in 1997 - 1999 at Research Center for Oceanography-LIPI. Biostimulation by using urea, modified customblen and oleophilic as fertilizer has been tested on oil-polluted sea water column by mesocosm approaches in field experiment (Ruyitno, 1997). Bioaugmentation has also been studied by using single culture and consortium local bacteria with the same approaches (Darmayati, 1999).

Improvement on bioremediation studies Indonesia has been in occurred bv involvement of molecular science since 2005. LIPI-NITE collaboration research after conducted. Biostimulation studies were done from laboratory scale and microcosm scale to field experiment (sand column). These were stressed on bioremediation in sand beach environment. It was different from the previous experiment which concentrated on the water column. The present studies showed that "Osmocote" was the best slow release fertilizer compared to other studies. Besides that the study revealed the microbial succession community during the bioremediation processes, the proper amount of fertilizer and the period of fertilizer application should be suitable and, also the impact on microalgae growth (Darmayati, 2009).

Other issue on biostimulation is bioactivator, a complex mixture that functions as biostimulans for the performance of specific bacteria to degrade petroleum hydrocarbons entering into marine environment. Study on oil natural biodegradation and development of bioactivator to overcome oil pollution in the sea has been conducted at University of

Hasanudin (Noor, 1999). The formula, hitherto named Bioactivator UH-1 (BUH-1), has been used for studies on petroleum hydrocarbon degradation in marine environment.

Bioaugmentation studies are still research. Several selected ongoing hydrocarbonoclastic bacteria have been tested on their capability to degrade oil and certain toxic compounds, also studied on their growth characteristic and on formulation of robust consortium bacteria. For this time being, bioaugmentation and biostimulation experiments are conducting in vitro by using oil contaminated sediment from Pari Island. For the near future, this experiment will be done on oil-contaminated beach for pilot plan scale.

## **FUTURE RESEARCH DIRECTION**

on these facts, bioremediation Based technology is promising to apply in a certain marine environment in Indonesia, especially for sandy beach and may be for mangrove forest. However, to have a manual or guidelines on conducting bioremediation in marine environment is not easy and still many steps have to be done. Several aspects concerning with this study such as the diversity of polluted sites characteristic, oils characteristic, oceanographic conditions and engineering has to be studied comprehensively. Besides, some trials and errors need to be passed to confirm the result, to identify and to overcome the limitation and a real need, to ensure that the discoveries in the laboratory can be successfully applied in the field.

### **CONCLUDING REMARKS**

As a conclusion (1) Indonesia need 'green' methods/techniques to control oil pollution, not only in soil environment but also in marine environment; (2) Many steps have to be done to develop bioremediation technology for marine environment; and (3) Collaboration research intra and inter disciplines/institutes/countries will accelerate achievement on oil pollution control.

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